techniques, to investigate the macromolecular structure of proanthocyanidins from several plant-derived foods. In fact, a whole series of novel cyclic B-type 4-, 5- and 6-mer proanthocyanidins could be distinguished from their isomeric non-cyclic A-type analogues by isotopic exchange mass spectrometry only.

Novel Aspect

A mass spectrometric method based on hydrogen/deuterium exchange for the resolution of novel cyclic isomeric proanthocyanidins in wine and plant-derived foods is presented.

References

1. Longo E., Rossetti F., *et al.*, *Journal of the American Society for Mass Spectrometry*, 29(4), 663 (2018).
2. Jouin A., Rossetti F., Teissèdre P. *et al.*, *10th IVAS Symphosium*, *Salamanca (Spain)*,160 (2017).
3. Jeffery D., Mercurio M.D., Herderich J. *et al.*, *Journal of Agricultural and Food Chemistry*, 56(8), 2571 (2008).

ThOr.05-5 *MALDI-TOF MASS SPECTROMETRY IDENTIFICATION AND DETECTION OF RELEVANT PATHOGENS IN BEEF CATTLE*

Gisele Bacanelli [(1)] - ***Cynthia Mantovani*** [(1)] - ***Anna Letícia Louzan*** [(1)] - ***Taynara Pasquatti*** [(2)] - ***Daniele Bier*** [(2)] - ***Gracia Rosinha*** [(3)] - ***Flabio Araujo*** [(3)] - ***Newton Verbisck*** [(3)]

[(1)] *Federal University of Mato Grosso do Sul, UFMS, Campo Grande* - [(2)] *Dom Bosco Catholic University, UCDB, Campo Grande* -
[(3)] *Embrapa Beef Cattle, Animal Health, Campo Grande*

Keywords: MALDI-TOF, Mycobacterium, Brucella, Salmonella, Beef Cattle.

Introduction

Mycobacterium bovis, *Brucella abortus* and *Salmonella* species account for significant economic losses in cattle production worldwide, besides being public health threats. We have established MALDI-TOF (Matrix Assisted Laser Desorption Ionization – Time-of-Flight) mass spectrometry with modified protein extraction methods and improved the presently available reference spectra libraries, in order to effectively detect those pathogens from bovine samples.

Methods

Bacterial isolation from bovine tissue and lesions followed microbiological standard methods. Heat inactivated mycobacteria from solid media were mechanically disrupted as designed herein. *Brucella* and *Salmonella* were grown in liquid media and processed as described [1]. Mass spectra acquired with alpha-cyano-4-hydroxycinnamic acid on Autoflex III Smartbeam underwent microorganism identification carried out on MALDI Biotyper software (Bruker Daltonik).

Results

Analysis of mycobacterial isolates revealed that sixty-three classified as *Mycobacterium bovis*, two as *Gordonia sputi* and one as *M. nonchromogenicum*. These results were confirmed by Polymerase Chain Reaction (PCR) specific for *M. bovis* and 16S rDNA sequencing (100% concordance).

Brucella abortus, *B. suis*, *B. ovis* and *B. canis* mass spectra profiles, generated as reference strains, were clearly different and allowed field samples identification. Amongst twelve isolates with gender specific PCR positive, only two were confirmed as *Brucella* by MALDI-TOF, indicating possible contaminations during cultivation. We also tested if *Brucella* fingerprint could be detected directly from tissue lesion homogenates, lacking any microbiological culture, which was successfully demonstrated.

Ninety-six isolates from bovine carcasses were biochemically tested for *Salmonella* and species were identified by MALDI Biotyper analysis. Four isolates showing inconclusive results in biochemistry actually belonged to *Citrobacter* and *Proteus* genera [2].

Conclusions

The cell processing method established here provides reliable *Mycobacterium* identification at species level, with scores that safely allow *M. bovis* and *M. tuberculosis* distinction of isolates from solid media culture. We have discriminated *Brucella* species and could detect this pathogen directly from bovine tissue lesions, skipping bacterial cultivation step. In addition, MALDI-TOF enabled *Salmonella* identification at species level with serovar indicative.

Novel Aspect

Distinction of Mycobacterium Tuberculosis Complex members at species level and detection of *Brucella* directly from infected tissue by MALDI-TOF mass spectrometry.

References

1. Freiwald A. & Sauer S., *Nature Protocols*, 4, 732 (2009).
2. Bier D., Tutija J., Pasquatti T., Oliveira T., Araujo F., Verbisck N., *Pesquisa Veterinaria Brasileira*, 37, 1373 (2017).

ThOr.06 – Lipidomics

Chairs: Gavin Reid, Donatella Caruso

Thursday

ThOr.06-1 Keynote: *LIPIDOMICS ANALYSIS OF CELLS AND TISSUES IDENTIFIES THERAPEUTIC TARGETS*

Michael Wakelam [(1)] - ***An Nguyen*** [(1)] - ***Qifeng Zhang*** [(1)] - ***Roberto Solari*** [(2)]

[(1)] *Babraham Institute, Signalling, Cambridge, United Kingdom* - [(2)] *Imperial College, Imperial College, London, United Kingdom*

Keywords: Mass spectrometry, lipidomics, pathway analysis, colorectal cancer, rhinovirus

Introduction

Theoretically mammalian cells can contain many thousands of individual lipid molecular species, whilst it is unlikely that all are present in a single cell, lipidomic experiments have demonstrated the presence of more than a thousand species. The integrated regulation of changes in lipid species that occur that can regulate cell functions, including signaling and metabolism, highlights the need for bioinformatics analysis to fully interpret lipidomics data.

Methods

We have adopted two experimental systems: lipids extracted from colorectal tumour tissue and rhinovirus-infected human bronchial epithelial cells, were identified by LC-MS/MS. Pathway analysis of lipid metabolising enzymes coupled to network optimising Prize-collecting Steiner tree problem methodology was adopted to identify key enzymatic changes which could be considered as therapeutic targets.

Results

Lipidomic analysis that compared colorectal tumour tissue of defined stage with matched normal tissue showed changes in more than 700 lipid species, this also demonstrated changes in acyl chain saturation and chain length accompanying tumour progression. In the rhinovirus-